SOYBEAN NODULATION RESPONSES TO DIFFERENT LIGHT ENVIRONMENTS OF THE SHOOT

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ABSTRACT

Spectral balance (quality) of canopy light influences many aspects of plant growth and development via its effect on phytochrome, and spectral balance in the seedling establishment zone is affected by reflection from plants, soils, or crop residues on soil surfaces. Soybean [Glycine max (L.) Merr.] growth responses to spectral balance change associated with either row orientation or spacing is particularly pronounced when strain USDA 110 of Bradyrhizobium japonicum is the inoculum. Soybean seedling growth and nodulation are also affected by soil surface color via influence of spectral composition of reflected light. Nodulation and root growth are greatest on plants grown over white surfaces. Total seedling dry matter is influenced most by PPFD, and stem length and nodulation are highly correlated to FR/R ratio and quantity of blue light. The FR/R ratio also affects the speed with which the autoregulation of nodulation on one side of a split-root system blocks nodulation on the other. Complete blockage occurs quicker with a high FR/R ratio. Thus, the interaction of the B. japonicum strain and the light environment of the shoot are important to nodulation and productivity of soybean.

INTRODUCTION

Dinitrogen fixation and the soybean-B. japonicum symbiosis are of considerable agricultural importance, particularly in geographic areas with low-nitrogen soils. Although considerable work has been directed toward improvement of B. japonicum strains, additions of new inoculants to field-planted soybean frequently do not increase the nodular occupancy of the introduced strains (Hunt et al., 1985a). These inconsistent results can be partly explained by B. japonicum and plant interaction with environmental variables such as drought, heat, and soil acidity (Keyser and Munns, 1979; Hunt et al., 1981; Munevar and Wollum, 1981). Recently, Hunt et al. (1985b) showed that the response of soybean seed yield to row orientation was influenced by the strain of B. japonicum used in the inoculation. They postulated that the far-red/red (FR/R) ratio of canopy light was involved, since they had previously shown in a controlled environment study that FR/R ratio in the soybean shoot environment regulated (via phytochrome) photosynthate partitioning between shoot and root as well as nodule mass and number (Kasperbauer et al., 1984). These changes in the spectral composition of canopy light have been associated with spectral distribution of reflected light from neighboring plants (Kasperbauer, 1987).

Light, of course, can be reflected from many surfaces. Soil surface and plant residue colors were recently recognized as contributors to variations in seedling light environment (Kasperbauer and Hunt, 1987). Differences in light reflected from black, red, and white surfaces were sufficient to cause very dramatic and consistent differences in stem length of southern pea [Vigna unguiculata (L.) Walp.] even though soil temperatures below the black and white surfaces were within 1^{o}C of each other.

Variations in amount of nodulation associated with canopy light spectral composition is likely expressed in the autoregulation of the nodulation process. Kosslak and Bohlool (1984) showed that nodulation of one side of a split-root system of soybean would suppress nodulation of the other side if inoculation of the second side was delayed a few days. They also showed that the effect was enhanced or diminished by the length of day and by photosynthetic photon flux density (as regulated by a shade cloth). They interpreted the response to be due to differences in photosynthate production levels. However, day length responses also involve the phytochrome system (Borthwick, 1972). Additionally, other workers (Andersen et al., 1985) reported that under field conditions, green shade cloth increased the FR/R ratio of penetrating light by about 5% and influenced plant growth. The purpose of our report is to assess the effects of canopy light composition on soybean nodulation.

METHODS AND MATERIALS

Greenhouse Studies

Experiments were conducted to determine effects of reflected light from various soil surface colors on soybean seedling growth. Pre-germinated Braxton soybean seeds were planted when the radicles were approximately 5 mm in length, inoculated with 10^8 cells of B. japonicum (USDA3I1B110), and grown in one-L containers that were attached below the colored surfaces. Containers were filled with horticultural-grade vermiculite and watered as needed with 0.25-strength N-free nutrient solution. Temperatures within the containers were measured at 1-min intervals with copper-constantan thermocouples and a Campbell CR7 Datalogger.[1] Upwardly reflected light was measured 10 cm above the surface at 5-nm intervals from 400 to 800 nm with a LiCor LI-1800 spectroradiometer with the light collector on a 1.5-m fiber optic probe. Upwardly reflected light values were expressed as the percentages of direct sunlight at each measured wavelength. Spectral irradiances at 735 nm and 645 nm were used to calculate the FR/R ratios because these wavelengths are the FR and R phytochrome action peaks, respectively, in green plants (Kasperbauer et al., 1964). The PPFD, FR/R photon ratio, and blue light quantity in upwardly reflected light 10 cm above the variously colored surfaces were measured. This height above the soil is important because it is in the seedling establishment zone. Plants were sampled, four weeks after planting. Details that differed between experiments are outlined below.

Experiment I. White, brick-red, near-black and oat straw residue-covered near-black soils were the reflective surfaces. The soil containers were 2.5-cm deep plywood boxes whose bottoms were covered by 122 x 122 x 2-cm styrofoam insulation panels. Four 2-cm holes were drilled in the styrofoam and box bottoms to allow passage of plants through 2-cm diameter plastic pipes that extended from the container of vermiculite below the styrofoam panel through the panel and the layer of soil. The soil covered with dry oat straw simulated an 85% crop residue cover, as frequently occurs in conservation tillage.

Experiment II. The effects of two levels of reflected blue light (400 to 500 nm) on soybean seedling growth were studied in this experiment by the use of yellow and light blue fabrics covering the surface of the panels. The two fabrics reflected almost identical PPFD and FR/R ratios. However, about 75% of

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1 Mention of a product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture and does not imply its approval to the exclusion of other products that may also be suitable.

the blue in direct sunlight was reflected from the blue surface while only 15% of the blue in direct sunlight was reflected by the yellow surface.

Controlled Environment
A split-root (sides A & B) study with soybean (cv Lee) and USDA strain 3Ilb110 *B. japonicum* was conducted to assess the effect of R (low FR/R ratio) and FR (high FR/R ratio) light treatments of shoots on the timing of *B. japonicum*-induced suppression of root nodulation. A modified split-root system as described by Singleton (1983) was used. Plants were grown at 25°C in a growth-chamber with 12-h daily photosynthetic periods that ended with 5-min exposures to R or FR light as described by Kasperbauer and Hamilton (1984). More detail can be found in (Hunt et al., 1987,1989)

RESULTS AND DISCUSSION

Variation in canopy light associated with row orientation and the attendant reflection of FR light from neighboring plants cause differences in both the growth and seed yield of soybean. These conclusions have been clearly documented in Kasperbauer et al. (1984), Hunt et al. (1985b), and Kasperbauer (1987) as well as by Kasperbauer and Hunt (these proceedings). These studies focus on the changes in FR/R ratio in light caused by reflection from neighboring plants, but canopy light is also effected by light reflected from the soil surface or crop residues (Kasperbauer and Hunt, 1987).

Data in Table 1 show differences in the spectral composition of light reflected from different soils. Plants grown over white surfaces developed the shortest stems, the most lateral roots and nodules, and the smallest shoot/root dry

Table 1. Upwardly reflected light, shoot and root growth, and nodule characteristics of soybean seedlings grown over different colored soils and oat straw residue (Hunt et al., 1989).

Light and plant characteristics	Soil surface color				LSD (0.05)
	White	Red	Black		
			Bare	Residue	
Upwardly reflected light 10-cm above surface[a]					
PPFD, $\mu mol\ m^{-2}s^{-1}$	129	56	30	75	-
Blue, % reflected	38	7	7	9	-
FR/R, ratio	0.85	0.91	0.94	1.03	-
Plant					
Stem length, mm	127	138	141	159	14
Stem weight, mg	137	120	120	136	15
Lateral root, mg	220	151	161	156	41
Total root, mg	293	213	222	217	36
Lateral nodule, mg	29	17	17	19	11
Shoot:Root weight ratio	1.48	1.69	1.57	1.75	0.22

[a]PPFD = photosynthetic photon flux density, FR/R = ratio of photons received at 735 nm divided by photons received at 645 nm, Blue = the amount of reflected blue light (400 to 500 nm) expressed as a percentage of incident blue light.

matter ratios. Since the amount of blue light and the FR/R ratio of light received by the shoots are involved in regulation of nodulation, an experiment was conducted to assess the impact of blue light quantity when the total PPFD and FR/R ratio were reasonably constant. Data in Table 2 show that the amount of blue, in addition to the FR/R ratio, in reflected light contributed to these morphogenic effects. It was concluded that the surface color of soil can substantially alter the growth and nodulation of soybean seedlings even though root zone temperatures under various surface colors are similar.

Table 2. Characteristics of soybean seedlings and upwardly reflected light over surfaces that reflected the same PPFD and FR/R ratios but different amounts of blue (400 to 500 nm) light (Hunt et al., 1989).

Characteristic	Reflected blue light		Sig*
	Low	High	
Soybean seedlings			
Stem length, mm	106	99	*
Root wt, mg/plant	125	138	*
Nodule wt, mg/plant	11	18	*
Upwardly reflected light 10-cm above surface[a]			
PPFD (μmol $m^{-2}s^{-1}$)	345	365	NS
FR/R (photon ratio)	0.89	0.87	NS
Blue (% of incident blue)	15	75	*

*indicates means differ at P=0.05 by paired comparisons.
[a]PPFD, FR/R and Blue as described in footnote of Table 1.

Whether light spectral changes received by the growing shoots resulted from canopy geometry or from soil surfaces, it is important to know if the timing as well as the amount of nodulation can be affected. The split-root technique offers a good method of assessing impact of an altered FR/R ratio in canopy light on the timing of nodulation. The technique was used in a controlled environment study. When side B was inoculated 2 to 4, or more, days after side A, nodulation and nodule mass on side B were significantly less than on side A. Number of nodules formed on roots of either side A or B were lower when shoots were treated with FR (a high FR/R ratio) than with R (a low FR/R ratio). Therefore, total suppression of nodulation on side B occurred more rapidly with FR than R light treatment. When FR was followed immediately by R, plants responded similarly to the R-only treatment for nodule weight as well as nodule number (Table 3). These data show that the timing of suppression of secondary nodulation on the roots can be affected by spectral balance of light received by the shoots, and the photoreversible nature of the effects of R and FR light strongly indicates phytochrome involvement.

Table 3. Effect of inoculation time and R and FR treatment of shoots on nodule dry mass for soybean grown in a split-root system (Hunt et al., 1987).

Inoculation schedule	Light treatment					
	R		FR		FR,R	
	Side		Side		Side	
	A	B	A	B	A	B
	mg/plant[a]					
A_0B_0[b]	9±2*	8±1*	4±1*	5±1*	5±2*	6±3*
$A_{6-9}B_{6-9}$	8±2*	7±2*	6±2*	6±2*	9±2*	6±2*
A_0B_{2-4}	14±2*	4±1*	6±2*	2±1*	6±0*	3±2*
A_0B_{4-6}	24±4*	2±1*	6±2*	1±0	15±4*	2±2*
A_0B_{6-9}	29±5*	1±1	14±5*	0±0	21±6*	0±0
A_0B_9	26±6*	0±0	18±4*	0±0	15±4*	0±0
Noninoc	0±0	0±0	0±0	0±0	0±0	0±0

[a] Values are means ± SE.
[b] Inoculation Day-0 was 7 days after planting, after roots had extended into each side of the split-root system. A and B designate the two sides of the split-root system, and subscripts designate the inoculation time (days) after Day-0.
* Numbers within the same column are significantly different from the noninoculated control at $\underline{P}<(0.05)$ when compared by a single degree of freedom contrast. Numbers were transformed by Box-Cox transformation = 0.11 for homogeneity of variance before analysis (Box and Cox, 1964).

CONCLUSIONS

Nodulation of soybean grown in different populations, row widths, and row orientations as well as over different colored soil and crop residues is affected by altered amounts of blue and the FR/R ratio of light received by the growing shoots. These altered environmental conditions and the attendant physiological responses may be important in the interpretation of field as well as greenhouse and growth-chamber studies. Thus, it is important to consider the interaction of the *B. japonicum* symbiont, soybean plant, and canopy light environment for optimal production of soybean.

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